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EXAMINER	
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/685,837	SEIBLER ET AL.	
	Examiner	Art Unit	
	Anoop Singh	1632	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *17 September 2007*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1,5,6,8-12,14-24,26,27 and 30* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,5,6,8-12,14-24,26,27 and 30* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Applicant's amendment filed on September 17, 2007, has been received and entered. Claims 1, 14,-15 and 30 have been amended and claim 2-4, 7, 13, 25, 28 and 29 have been canceled.

A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on September 17, 2007 has been entered.

Claims 1, 5-6, 8-12, 14-24, 26-27 and 30 are pending.

Election/Restrictions

Applicant's election with traverse of the invention of group IV (27) filed October 24, 2005 was acknowledged. Applicant argument of examining method for gene knock down in a vertebrate (group 1) with elected group were found persuasive, therefore invention of group I and IV directed to vertebrate and method of gene knock down in a vertebrate were rejoined for the examination purposes.

Accordingly, a method for gene knock down in a vertebrate and vertebrate having stable integration at Polymerase II dependent locus, an expression vector comprising an shRNA construct under control of a ubiquitous promoter were examined in the instant application.

Claims 1, 5-6, 8-12, 14-24, 26-27 and 30 are under consideration.

Withdrawn-Claim Objections

The objection to claim15-17 as being of improper dependent form for failing to further limit the subject matter of a previous claim is withdrawn in view of amendments to the claims.

Claim Objections

Claim 1 is objected to because the term "homologous " is misspelled as in line 6 of the claim. Appropriate correction is required.

New- Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1, 5-6, 8-12, 14-24, 26 and 27 remain rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method of gene knockdown in a mouse genome at the *rosa26* locus, said method comprising introducing a reporter construct comprising shRNA in mouse embryonic stem cell by homologous recombination, wherein said shRNA and reporter constructs comprises a gene encoding Renilla (Rluc) and luciferase (Fluc) along with an adenovirus splice acceptor sequence and polyadenlylation signal placed downstream of the endogenous promoter of rosa26, and Fluc specific shRNA expressed under the control of H1 and U6 promoter and terminated by five thymidines; and microinjecting said mouse embryonic stem cell into mouse diploid blastocysts; and implanting the blastocysts comprising the mouse embryonic stem cell into pseudo pregnant mouse; allowing the resulting pregnant mouse to deliver viable chimeric offspring and a transgenic mouse produced by said method, wherein said transgenic mouse exhibits ~90% reduced luciferase activity in liver, heart, brain

and muscle, does not reasonably provide enablement for the method of gene knockdown in any other nonhuman vertebrate which comprises stably integrating at any Polymerase II dependent locus any other shRNA sequence. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

In determining whether Applicant's claims are enabled, it must be found that one of skill in the art at the time of invention by applicant would not have had to perform "undue experimentation" to make and/or use the invention claimed. Such a determination is not a simple factual consideration, but is a conclusion reached by weighing at least eight factors as set forth in In re Wands, 858 F.2d at 737, 8 USPQ 1400, 2d at 1404. Such factors are: (1) The breadth of the claims; (2) The nature of the invention; (3) The state of the art; (4) The level of one of ordinary skill in the art; (5) The level of predictability in the art; (6) The amount of direction and guidance provided by Applicant; (7) The existence of working examples; and (8) The quantity of experimentation needed to make and/or use the invention.

The office has analyzed the specification in direct accordance to the factors outlines in *In re Wands*. MPEP 2164.04 states: "[W]hile the analysis and conclusion of a lack of enablement are based on factors discussed in MPEP 2164.01(a) and the evidence as whole, it is not necessary to discuss each factor in written enablement rejection." These factors will be analyzed, in turn, to demonstrate that one of ordinary skill in the art would have had to perform "undue experimentation" to make and/or use the invention and therefore, applicant's claims are not enabled.

Claim 1 encompasses a method for constitutive and/or inducible gene knock down in a nonhuman vertebrate selected from a list consisting of mouse and fish, comprising an shRNA construct under control of a ubiquitous promoter and homologous sequence which integrates through homologous recombination at the polymerase II dependent locus of the genome of the nonhuman vertebrate. The

dependent claim 5 limits the polymerase II dependent locus selected from list of a group consisting of rosa26, collagen, RNA polymerase, actin and HPRT locus. Claims 6, 8-14 encompasses the expression vector of claim 1, which further contains functional sequences and group of promoters, which is either constitutive or inducible. The inducible promoter expression is a promoter consisting an operator sequence and the vertebrate of claim 1 is non-human vertebrate, which is further limited to either mouse or fish. Claims 15-19 encompass method of claim 1 wherein the vector is a either Pol III or Pol II dependent promoter driven shRNA construct suitable for integration into a Pol II dependent locus. Subsequent claim limit the Pol III promoter being either constitutive or inducible H1 or U6 promoter while Pol II promoter being inducible CMV promoter. Claims 20-24, 26-27 encompasses method of claim 1 which describes the shRNA segment comprising a stop and or polyadenylation sequence that is integrated at polymerase dependent locus of the vertebrate and ES cell of the vertebrate.

Claims 1, 5-6, 8-24, 26 and 27 are broad in scope. The following paragraph will outline the full scope of the claims: Claimed invention recites a method of gene knockdown in a nonhuman vertebrate selected from a list consisting from mouse and fish, wherein said vertebrate comprises stably integrated by homologous recombination at any polymerase II dependent locus, an expression vector comprising any short hairpin RNA (shRNA) constructs under control of any ubiquitous promoter. Since these claims are broad in scope, encompassing mouse or fish having stably integrated by homologous recombination at any Pol II dependent locus using any promoter, subsequently limiting to few ubiquitous promoter, the disclosure provided by the applicant, in view of prior art, must encompass a wide area of knowledge to a reasonably comprehensive extent. In other word each of those, aspect considered broad must be shown to a reasonable extent so that one of the ordinary skills in the art at the time of invention by applicant would be able to practice the invention without any undue burden being on such Artisan.

The specification broadly discloses the progression of RNA interference technology over the years (pages 1,2) and describes role of shRNA-mediated gene silencing in transgenic mice and rats. The invention is based in part of a method of using a expression vector comprising a short hairpin RNA construct under the control of ubiquitous promoter for gene knock down in a living organism (pp 3). Pages 4-6 provide short description of figures. Pages 7-14 of the specification disclose definition of terms, general description of ubiquitous promoter, expression vector and a general description of different shRNA sequence in tabular form. Pages 15-17 broadly discusses preferred embodiments of the method steps comprising of generating shRNA and construct for cell culture, luciferase measurement assay and generation of chimeric mice. Example 1: of specification teaches the firefly luciferase gene along with a splice acceptor sequence is inserted into first allele of rosa26 locus by homologous recombination in ES cells while shRNA and Renilla luciferase gene is inserted into second allele of rosa 26. Figure 7 shows the expression of the firefly luciferase in presence and absence of shRNA expression cassette. Example 2 shows shRNA expression cassette under control of U6 promoter containing tet operator sequence and a Renilla luciferase gene is inserted into first allele of rosa26 locus (figure 8 and 12), while the luciferase gene with a promoter and a tet repressor expression cassette is introduced into the second allele in ES cells. Luciferece activity is shown in presence and absence of doxycycline. Example 3 page 19 describes that NIH3T3 cells are transiently transfected with construct expressing the luciferase and tet repressor gene together with the shRNA construct containing tet operator sequence. Figure 11 shows the expression of luciferase in presence and absence of doxycycline. The specification discloses doxycycline inducible shRNA expression resulted ~80% inhibition in firefly luciferase activity in cells. Example 4 pages 19 show chimeric mice from rosa26/U6 and H1-ShRNA transgene. The data shows shRNA construct under the control of both U6 and H1 effectively repressed the firefly luciferase activity inmost organs (Figure 13B).

However, such broad disclosure does not demonstrate the information required by the Artisan to reasonably make and use mouse or fish with gene knock down by using an expression vector comprising any short hairpin RNA (shRNA) construct under control of a any ubiquitous promoter integrated by homologous recombination at any polymerase II locus.

As a first issue, instant claims embrace stably integrating by homologous recombination an expression vector into a polymerase II dependent locus wherein the expression vector comprises any shRNA under control of any ubiquitous promoters and homologous sequence that integrates through homologous recombination at polymerase II dependent locus in the genome of the fish or mouse. In the instant case, Applicant has exemplified a method to gene suppression of Renilla (Rluc) and luciferase. It is noted that none of the examples demonstrate specific *in vivo* gene silencing of any other gene encoding any other DNA segment. The specification teaches a method of shRNA within the vector of the invention comprises at least one DNA segment A-B-C wherein A is a 15 to 35, preferably 19 to 29 bp DNA sequence being at least 95%, preferably 100% complementary to the gene to be knocked down (e.g. firefly luciferase, p53, etc.); B is a spacer DNA sequence having 5 to 9 bp forming the loop of the expressed RNA hair pin molecule, and C is a 15 to 35, preferably 19 to 29 bp DNA sequence being at least 85% complementary to the sequence A (see para 40-44 of the specification). Thus, independent claims embrace a DNA segment of any length, however the specification does not support the use of the large DNA segment gene in an RNAi. In addition, prior and post filing art indicate specific nucleotide length requirements for RNAi to be effective. Elbashir et al (Genes Dev. 2001 15; 15(2):188-200, IDS) demonstrates that 21 and 22 mer act as act as guide RNAs for sequence specific mRNA degradation and therefore act most effectively in RNAi. Elbashir et al also describes that 30 bp dsRNA are ineffectively processed to 21-22nt RNA suggesting ineffective gene silencing by larger nucleotide sequence (page188 col. 2, para 2).

The specification further describes stably integrating, preferably at a polymerase II dependent locus of the living organism. The specification further discloses that polymerase II dependent loci include Rosa26 locus, collagen, RNA polymerase, actin and HPRT. Prior to instant invention, the unpredictability of attenuating /inhibiting expression of a target gene in cell by RNAi is evident in prior and post filing art. While it is recognized, that introduction of dsRNA that is targeted to a specific gene may result in attenuation /inhibition of the targeted gene, the degree of attenuation and length of the time attenuation is achieved in not predictable. Caplen et al (Gene 2000, vol. 252, 95-105, art of record) provide evidence of the unpredictability of dsRNA attenuation /inhibition of targeted gene in vertebrate cell *in vitro*. Transient transfection of dsRNA to the βgal transgene into 293 and BHK31 cells resulted in either no effect or a non-specific decrease in gene expression (pp102; Figure 7 A and B). Prior to instant invention, homologous recombination in embryonic stem cells has been widely used to produce mice carrying a single copy of the transgene integrated into a predetermined site inlcuding polymerase II dependent locus of the genome (Bronson et al., Proc. Natl. Acad. Sci. USA, 93(17:9067-72 (1996) art of record). However, art also teaches stably integrating at any polymerase II dependent locus would not necessarily yield predictable gene knock down. This is evident from expression of the low expression of the transgene. For instance, Hatada et al. demonstrated that the HPRT locus suppresses the activity of both, the haptoglobin gene promoter as well as the herpes simplex thymidine kinase promoter in several tissues of mice (Hatada et al., J. Biol., Chem., 274(2):948-55, 1999, IDS). Likewise, a human eNOS promoter-LacZ reporter gene placed in the Hprt locus was found to be inactive in hepatic vessels that otherwise express the endogenous eNOS gene (Guillot et al., Physiol. Genomics, Mar. 13, (2):77-83, 2000). Thus, it is clear from the cited art that, the several factors confounded the method for gene knockdown and integration of expression vector comprising shRNA into any polymerase II dependent locus the genome of the

nonhuman vertebrate that would not necessarily result in gene knockdown commensurate with full scope of the claims. It is noted that the unpredictability of a particular art area may alone provide reasonable doubt as to the accuracy of the broad statement made in support of enablement of claims. See *Ex parte Singh, 17 USPQ2d 1714 (BPAI 1991).* It is also well established in case law that the specification must teach those of skill in the art how to make and how to use the invention as broadly claimed. *In re Goodman, 29 USPQ2d at 2013 (Fed. Cir. 1994),* citing *In re Vaeck, 20 USPQ2d at 1445 (Fed. Cir. 1991).* An artisan would have to perform undue experimentation without reasonable expectation of success in order to make and use the invention commensurate with full scope of the claims.

As a second issue, claim 1 embraces a method for gene knockdown in nonhuman vertebrate selected from a list consisting of mouse or fish which comprises stably integrating by homologous recombination an expression vector into an polymerase II dependent locus of the genome of the nonhuman vertebrate, wherein said vector comprising a shRNA construct under control of any ubiquitous promote selected from group consisting of polymerase I-III dependent promoter. The specification contemplated that the expression vector of the instant invention is suitable for stable integration into the nonhuman vertebrate (see paragraph 32). The specification provides working example showing a method of gene knockdown by introducing a shRNA and reporter constructs in mouse embryonic stem cell by homologous recombination, wherein said shRNA and reporter constructs comprises a gene encoding Renilla (Rluc) and luciferase (Fluc) along with an adenovirus splice acceptor sequence and polyadenlylation signal placed downstream of the endogenous promoter of rosa26, and Fluc specific shRNA expressed under the control of H1 and U6 promoter (see example 1 and 4). However, specification fails to provide enabling disclosure with respect to obtaining ES cells from the fish or stably integrating at another polymerase II dependent locus as broadly embraced by the breadth of the claims. The specification has exemplified a method that embraces

an expression vector comprising shRNA for stable, rosa 26 locus dependent integration of the construct using embryonic stem cell. The art at the time of filing held that transgenic technology using ES cell was not predictable for any species of fish other than mouse. Since the specification discloses using mouse ES cells to produce transgenic mice via homologous recombination of targeting vectors in the ES cells, ES cells from various species of fish would be required as recited in the claims. However, prior and post filing art teaches that germline competent embryonic stem cells are not available in any other mammalian species other then mouse (Denning et al., Reproduction 126:1-11, 2003, Hochepied et al Stem Cells, 2004, 22, 441-447; abstract). It is noted that several years after filing of instant application, Hong et al (Methods Mol Biol. 2006;329:3-16) provided guidance with respect to obtaining medaka ES cells, however, it is generally known in the art that homologous recombination of a targeting construct had not been attempted with ES like cells available prior to filing of this application. In fact a recent report Alvarez et al (Mar Biotechnol . 2007 Mar-Apr;9(2):117-27) acknowledges the slow progress made in recent years and asserts that "there are promising achievements in homologous recombination and alternative avenues yet to be explored that can bring ES technology in fish to fruition" (abstract). This clearly shows that targeting gene by homologous recombination using fish ES cells was still evolving at the time of filing of this application. In the instant case, neither specification nor prior art provided any guidance with respect to targeting a construct for stably integrating by homologous recombination at a polymerase II dependent locus a construct comprising shRNA under the control of a promoter in any species of fish for the purposes of gene knockdown. Therefore, at the time of filing of this application, method of gene knockdown in could not have been accomplished for any species of fish other than mouse. The specification does not teach how to make knock down nonhuman vertebrate by shRNA for any other species other than mice or correlate making mice to making knockout fish of any species using the method as recited in

impendent claims. Therefore, the claims should be limited to mouse and method for gene knockdown in mouse as discussed in the previous office actions. An artisan would have to perform undue experimentation for effective reduction to practice without reasonable expectation of success, especially in view of the evidence that is contrary to the instant claims.

As a final issue, claim 27 recites a nonhuman vertebrate selected from a list consisting of mouse and fish comprising an expression vector comprising a short hairpin RNA (shRNA) construct under control of a ubiquitous promoter integrated at a polymerase II dependent locus. As recited instant claims do not require any specific phenotype for the claimed transgenic fish or mouse and specification dose not provides adequate correlation between any phenotype obtained in the transgenic mouse to the phenotype obtained in any species of fish. It is generally known that only mouse is routinely manipulated animal and prior art recognized that the phenotypes of transgenic mouse does not predict the phenotype in non-mice species. The state of the art at the time of filing uses the unpredictability of obtaining transgenic animals with a specific phenotype as summarized by the references of Keri et al., (Proc Natl Acad Sci U S A. 2000; 97(1):383-7) show that elevated levels of lutenizing hormone in transgenic can result in different reproductive system abnormalities including ovarian tumors. Similarly, Carmell et al failed to produce any distinct phenotype, while shRNA, constructs directed against seven known targets were introduced via standard trasngenesis (Carmell MA Nat Struct Biol. 2003; 10(2): 91-92, art of record). Thus it is clear from the cited arts that at the time of filing, the resulting phenotype of a gene knockdown resulting from methods routinely used for integrating shRNA in the genome of nonhuman vertebrate was considered unpredictable. In the instant case, specification as filed does not provide any specific information about resulting phenotype of the nonhuman vertebrate consisting of mice and fish. It is noted that the specification merely recites the luciferase activity in different organ, however it

dose not provide any specific information for practicing the claimed invention commensurate with the full scope of the claim. Holschneider et al. (Int J Devl Neuroscience, 2000, 18: 615-618) state that single genes are often essential in a number of different physiological processes. Hence, deletion of an individual gene may prove so drastic or so widespread as to create an amalgam of phenotypes whose interpretation becomes confounded by the interaction of various new physiologic changes (pp 615). Holschneider et al discuss various factors that contribute to the resulting phenotype of transgenic mice, including compensatory system that may be activated to mask the resulting phenotype; these compensatory changes may be due to differential expression of another gene, which may be regulated by the downstream product of the deleted gene. Thus, at the time of filing, the resulting phenotype of a knockout nonhuman vertebrate was considered unpredictable and it was confounded by multiple compensatory pathways. The specification does not teach any transgenic nonhuman vertebrate comprising any disruption that would result in expected phenotype. An Artisan of skill would need to perform further research upon the nonhuman vertebrate obtained by the process disclosed in the instant application in order to determine the correlation between the transgene knockdown and the observed phenotypes or effect. In absence of any specific teaching an artisan of skill would have to perform undue experimentation to make new invention in the field to make use of the invention. An artisan would have to perform undue experimentation to determine the appropriate elements that would specifically express genus of different genes in the nonhuman vertebrate showing expected phenotype. Absent of evidence to the contrary, it is not clear that resulting phenotype of a nonhuman vertebrate comprising genus of shRNA of known or unknown biological function particularly in view of unpredictably expressed in the art. An artisan would not know how to use resulting nonhuman vertebrate selected from a list consisting of mouse or fish made by the method described in the

specification and therefore would require undue experimentation to determine how to use the resulting transgenic nonhuman vertebrate.

In view of lack of teaching or guidance provided by the specification with regard to an enabled method for gene knockdown in any vertebrate comprising a disruption in gene using any shRNA, construct comprising different constitutive or inducible promoter, and shRNA sequence and the lack of teaching or guidance provided by the specification to overcome the art recognized unpredictability of disruption of a particular gene, promoter and locus and the resulting phenotype and absence of any correlation between disruption and its phenotype, for the specific reason cited above in the office action. It would require undue experimentation for an Artisan to make and use the claimed invention and/or working examples demonstrating the same, such invention as claimed by the applicant is not enabled for the claimed inventions commensurate with the full scope of the claims.

Withdrawn-Claim Rejections- - 35 USC § 112

Claims 15-17 rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention is withdrawn in view of amendments to the claims.

Maintained-Claim Rejections- 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 1, 5-6, 8-12, 14-24 and 26 remain rejected under 35 U.S.C. 112, second paragraph, as being incomplete for omitting essential steps, such omission amounting to a gap between the steps. See MPEP § 2172.01. Instant claims are directed to a method for gene knockdown comprising stably integrating by homologous recombination into a polymerase dependent locus an expression vector comprising shRNA construct, but the does not set forth any steps involved in method that would facilitate homologous recombination, it is unclear what method /process applicant is intending to encompass. The omitted steps are: homologous recombination is achieved by integrating expression vector in ES cell of the nonhuman vertebrate. Furthermore, independent claim merely recites a method of gene knock down by administering the vector by homologous recombination without any active, positive step linking to the preamble indicating whether method actually results in gene knock down phenotype. Claims 5-6, 8-12, 14-24 and 26 and directly or indirectly depends on independent claim. Appropriate correction is required.

In absence of any argument, claims 1, 5-6, 8-12, 14-24, 26-rejected under 35 U.S.C. 112, second paragraph is maintained for the reasons of the record . It is emphasized that independent claim do not recite any positive method step linking to preamble, therefore, it is unclear if method actually resulted in gene knock down in any nonhuman vertebrate.

New-Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham* v. *John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

Claims 1, 5-6, 8-10, 14-16, 18, 20-24, 26-27 and 30 are rejected under 35 U.S.C. 103(a) as being unpatentable over McCaffrey et al., (Nature, 2002 Vol. 418, 38-39) or Beach et al. (US patent Publication no. 2003/0084471, dated 5/1/2003, effective filing date 1/22/2002) and Bronson et al (Proc Natl Acad Sci U S A 1996; 93:9067–9072).

Note: Instant 103(a) rejection is applied to the extent claims are directed to a method of gene knockdown by stably integrating by homologous recombination an expression vector comprising shRNA under the control of a ubiquitous promoter. It is emphasized that none of the method steps require any specific expression level that results in gene knock down. The claims are limited to a method comprising (1) stably integrating by homologous recombination an expression vector comprising shRNA construct under the control of a ubiquitous promoter and homologous sequence which integrates through homologous recombination at polymerase II dependent locus. It is emphasized that instant rejection is to the breadth of the claims.

McCaffrey et al teach a method of gene knock down in a transgenic mouse comprising an expression vector comprising shRNA under the control of ubiquitous promoter (see abstract). McCaffrey et al teaches delivering an expression vector comprisng small-hairpin RNAs (shRNAs) that is expressed *in vivo* from DNA templates using RNA polymerase III promoters inhibiting the luciferase expression

by up to 98% (pp38, Figure 1 C-D and pp39 2nd paragraph). McCaffrey teaches shRNA that comprises at least one DNA segment A-B-C wherein A is a 15 to 29 bp DNA sequence with at least 100% complementarily to the gene to be knocked down; B is a spacer DNA sequence having 5 to 9 bp forming the loop of the expressed RNA hairpin molecule, and C is a 19 to 329 bp DNA sequence and further comprises a poly A sequence meeting the limitation of claims 20-24 (see the supplementary information). Although, McCaffrey et al taught a method of gene knockdown in a mouse, but differed from claimed invention by not disclosing stably integrating the expression construct in a polymerase II dependent locus.

Beach et al disclose that the double-stranded structure may be formed by a single self-complementary RNA strand or two complementary RNA strands. RNA duplex formation may be initiated either inside or outside the cell. Inhibition is sequence-specific in that nucleotide sequences corresponding to the duplex region of the RNA are targeted for genetic inhibition (pp4, paragraph 52). Beach et al teach the length of the dsRNA is at least 20, 21 or 22 nucleotides in length, e.g., corresponding in size to RNA products produced by Dicer-dependent cleavage. In certain embodiments, the dsRNA construct is at least 25, 50, 100, 200, 300 or 400 bases (pp13, paragraph 16). Beach et al disclose that the dsRNA construct may be synthesized either *in vivo* or *in vitro*. RNA can be derived from an expression construct (pp 13,14; paragraph 168). The invention also discloses strategy for stable expression of dsRNA in cultured mammalian cells (Figure 27, paragraph 78). Beach et al disclose generating several types of short dsRNAs corresponding to the coding region of firefly or Renilla luciferase (pp22; paragraph 246). Beach et al demonstrates that short hairpins encoded on a plasmid are effective in suppressing luciferase gene expression (Figure 42) *in vivo*. DNA oligonucleotide encoding 29 nucleotide hairpins corresponding to firefly luciferase were inserted into a vector containing the U6 promoter. Beach further discloses that one of skill can choose from amongst a range of vectors to either transiently or stably express a short

hairpin. Beach et al also disclose non-limiting examples of vectors and strategies to stably express short dsRNAs using U6 and H1 promoters (pp23; paragraph 252; Figures. 43-45). It is noted that Beach et al also disclose that promoters/enhancers that may be used to control the expression of the targeted gene in vivo may include cytomegalovirus (CMV) promoter (see para. 147). Beach et al teach and claim a non-human transgenic vertebrate selected from a list consisting from mouse or fish (see page 12, para. 154) having germline and/or somatic cells comprising a transgene encoding a dsRNA construct (pp 26, claim 28 and pp 2 paragraph 52) that includes rodent (pp12, paragraph 154). Beach et al also demonstrates that a short hairpin is highly effective in specifically suppressing gene expression of firefly or Renilla luciferase (Example 6). However, Beach et al do not explicitly teach how an expression vector integrates through homologous recombination at polymerase II dependent locus.

Prior to instant invention, Bronson describes transgenic mice made by pro nuclear injection of DNA as an effective method of achieving expression of exogenous DNA sequences for many purposes, including over expression, mutant analysis, promoter analysis (see page 9067, column 1, para 1). Bronson also describes problems associated with DNA incorporated into the mouse germ line using this method includes random integration and unpredictable copy numbers. This random integration often also presents profound effect on expression of the transgene resulting in altered phenotype of the mouse (see page 9067, col.1, para. 1). It is noted that Bronson provided advantages of targeting a single copy of a transgenic sequence to a chosen location in the genome such as HPRT over random integration of construct. He discloses many advantages of targeting at specific locus including the ability to control copy number, the ability to insert the transgene into regions of chromatin compatible with a desired developmental and tissue-specific expression. It is noted that Bronson et al emphasize that targeted transgenes provide a more efficient and informative means of securing and comparing the expression of various

transgenic sequences than is available with current transgenic procedures. Bronson also taught homologous recombination in murine ES cells to generate mice having a single-copy of a transgene inserted at a chosen site in the genome (see page figure 2 and page 9068, column 2, para 3). Specifically, Bronson et al disclose a method wherein a single copy murine bcl-2 cDNA driven by either a chicken beta-actin promoter or a human beta-actin promoter has been inserted immediately 5' to the HRPT locus by a directly selectable homologous recombination event (see the abstract and figure 2). However, Bronson et al do not teach expressing shRNA in a specific locus.

It would have been obvious for one of ordinary skill in the art at the time of invention to modify the method of gene knock down disclosed by McCaffrey or Beach to include the shRNA expression cassettes that are flanked by homology regions for the polymerase II dependent locus (HPRT) as disclosed by Bronson to stably integrate by homologous recombination in ES cells to generate nonhuman vertebrate having a single-copy of a transgene inserted at a chosen site in the genome. Bronson provided guidance by emphasizing that the use of a chosen site for a single copy of a transgene avoids many of the problems associated with randomly inserted transgenes (see page 9072, col. 1, last paragraph). It would have been prima facie obvious for one of ordinary skill in the art to make transgenic nonhuman animal that comprises stably integrated expression vector comprising an shRNA into a specific locus such as HPRT by homologous recombination as discussed by Bronson in order to more efficiently suppress the transgene expression for sustained period.

One who would practiced the invention would have had reasonable expectation of success because McCaffrey/Beach et al had already described a method for gene knockdown in a mice by transiently as well as stably expressing shRNA construct and it would have only required routine experimentation to modify the expression construct that are flanked by homology regions for the

polymerase II dependent locus as disclosed by Bronson. One of ordinary skill in the art would have been studied Bronson to combine the teaching of Beach/ McCaffrey because a method of gene knockdown in a mouse comprising a shRNA construct under control of a ubiquitous promoter into a specific polymerase II dependent locus would have provided stable and sustained expression of short hairpin resulting in gene knockdown.

Therefore, the claimed invention would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention.

Claims 1, 5, 27 and 30 are rejected under 35 U.S.C. 103(a) as being unpatentable over Beach et al. (US patent Publication no. 2003/0084471, dated 5/1/2003, effective filing date 1/22/2002) or McCaffrey et al., (Nature, 2002 Vol. 418, 38-39, art of record); Bronson et al (Proc Natl Acad Sci U S A 1996; 93:9067–9072, art of record) as applied to claims 1, 5-6, 8-10, 14-16, 18, 20-24, 26-27 and 30 above, and further in view of Soriano et al (US patent 6,461,864, October 8, 2002, art of record).

The teaching of Beach et al or McCaffrey and Bronson et al have been discussed above and relied in same manner here. Although combination of Beach /McCaffrey and Bronson taught a method of stably integrate by homologous recombination an shRNA construct under the control of a promoter in polymerase II dependent locus (HPRT) but differed from claimed invention by not disclosing stably integrating into other polymerase II dependent locus such as Rosa26.

Soriano et al teach methods and vector constructs for the production of genetically engineered non-human animals, which ubiquitously express a heterologous DNA segment in Rosa 26 locus (abstract and claim 1). It is noted that Soriano describes targeting region as a portion of a targeting construct which becomes integrated into an endogenous chromosomal location following homologous recombination between a homology clamp and an endogenous gene locus, such as a

ROSA26, ROSA5, ROSA23, ROSA11, G3BP (BT5), or EphA2 gene locus sequence (column 3, lines 51-54). Soriano taught a method of targeting region that is flanked on each side by a homology clamp, such that a double-crossover recombination between each of the homology clamps and their corresponding endogenous gene sequences result in replacement of the portion of the endogenous gene locus by the targeting region. However Soriano et al differed from instant method by not disclosing using shRNA construct in rosa26 locus.

It would have been obvious for one of ordinary skill in the art at the time of invention to modify the method of gene knock down disclosed by McCaffrey/Beach to include shRNA expression cassettes that are flanked by homology regions for the polymerase II dependent locus (rosa26) by homologous recombination in ES cells to generate nonhuman vertebrate having a single-copy of a transgene inserted at a chosen site in the genome. Bronson provided guidance by emphasizing that the use of a chosen site for a single copy of a transgene avoids many of the problems associated with randomly inserted transgenes (see supra and page 9072, col. 1, last paragraph). The reference of Soriano provided guidance with respect to ubiquitously expressed gene loci for use include Rosa 26, rosa5 and others (see col. 3, lines 49-54). It would have been obvious for one of ordinary skill in the art to try a method of gene knock down in nonhuman vertebrate by modifying the shRNA expression cassettes under the control of the CMV/H1 or U6 promoter as disclosed by McCaffrey/Beach and then flanking by homology regions for the Rosa26 locus to stably integrate expression cassette comprising an shRNA under control of ubiquitous promoter into a specific locus such as HPRT/rosa26 as discussed by Bronson with reasonable expectation of achieving predictable result to more efficiently suppress the transgene expression. It is noted that several polymerase II dependent loci were known at the time of filing of this application and it would have required only routine experimentation to flank expression cassettes comprising

shRNA under the control of a promoter with the homology regions of other polymerase II dependent locus (See MPEP2144.04).

One who would practiced the invention would have had reasonable expectation of success because McCaffrey/Beach had already described a method for gene knockdown in a mice by random integration of the construct and it was routine to use express transgene in a chosen site to avoid many of the problems associated with randomly inserted transgenes as evidenced from the teaching of Bronson. Thus, it would have only required routine experimentation to modify the expression construct that are flanked by homology regions for the polymerase II dependent locus as disclosed by Bronson. One of ordinary skill in the art would have been studied Bronson to combine the teaching of Beach/ McCaffrey and Soriano because a method of gene knockdown in a mouse comprising a shRNA construct under control of a ubiquitous promoter into a specific polymerase II dependent locus that included hprt, rosa26 or any other endogenous loci would have provided stable and sustained expression of short hairpin resulting in gene knockdown.

Therefore, the claimed invention would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention.

Claims 11-12, 17 and 19 are rejected under 35 U.S.C. 103(a) as being unpatentable over Beach et al. (US patent Publication no. 2003/0084471, dated 5/1/2003, effective filing date 1/22/2002, art of record) or McCaffrey et al., (Nature, 2002 Vol. 418, 38-39, art of record); Bronson et al (Proc Natl Acad Sci U S A 1996; 93:9067–9072, art of record) and Soriano et al (US patent 6,461,864, October 8, 2002) as applied to claims 1, 5-6, 8-10, 14-16, 18, 20-24, 26-27 and 30 above, and further in view of Ohkawa et al (Hum Gene Ther. 2000; 11 (4): 577-85; IDS).

The combined teachings of or McCaffrey/Beach, Bronson and Soriano have been discussed above and are relied upon in same manner. However, none of the reference explicitly teaches an inducible system.

Ohkawa et al teach several constructs composed of the human U6 snRNA promoter and sequences derived from the gene for the tetracycline operator of a prokaryotic tetracycline resistance transposon (abstract). Ohkawa also disclose that expression of the promoter of the human gene for U6 snRNA that contains tet O sequences between the PSE (Figure. 1 and 2) and a TATA box could be be efficiently repressed in cells with the Tet repressor and that this repression can be reversed by tetracycline. Ohkawa et al used this expression system to control the function of an antisense RNA for a fusion gene composed of genes for epidermal growth factor receptor (EGFR) and green fluorescent protein (GFP) and expression of this chimeric gene could be efficiently and rapidly inhibited by tetracycline. However Ohkawa et al do not teach a method to gene knockdown in a nonhuman vertebrate.

It would have been obvious for one of ordinary skill in the art at the time of invention to modify the construct and method disclosed by McCaffrey/Beach to include inducible promoters for shRNA construct wherein operator sequence consist tet as disclosed by Ohkawa. One of ordinary skill in the art would be further motivated to include this construct in a specific locus by homologous recombination in ES cells to generate nonhuman vertebrate having a single-copy of a transgene inserted at a chosen site in the genome wherein transgene could be regulated by tetracyline. Ohkawa provided the provided motivation by showing that tet based system could control the expression of transgene, while Bronson emphasized the use of a chosen site for a single copy of a transgene avoids many of the problems associated with randomly inserted transgenes (see page 9072, col. 1, last paragraph). Furthermore, Bronson and Soriano provided guidance with respect to different endogenous loci including Rosa 26 locus. The person of ordinary skill in the art would have been studied Bronson to make transgenic nonhuman animal comprising stably integrated expression vector comprising an shRNA under the control of ubiquitous promoter into a specific locus such as ROSA26 or HPRT.

One who would practiced the invention would have had reasonable expectation of success because McCaffrey/Beach had already described a method for gene knockdown in a mice by random integration. It would have only required routine experimentation to combine the teaching of McCaffrey/Beach, Bronson, Ohkawa and Soriano because a method of gene knockdown in a mouse comprising a shRNA construct under control of a tet based inducible promoter into a specific ROSA26/HPRT locus would have provided stable and sustained regulated inhibition of transgene.

Therefore, the claimed invention would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention.

Response to Arguments

As an initial matter it is noted that examiner has included new references in 103 rejections, thus, instant response is limited to the previously applied references of Beach; Bronson et al and Soriano et al.

Applicant's arguments filed September 17, 2007have been fully considered but they are not persuasive. Applicants argue that Soriano et al relate to ubiquitous expression of a heterologous gene inserted into rosa26 locus by homologous recombination, wherein rosa26 promoter drives the transgene expression via splice acceptor sequence. Applicants' assert that Soriano does not relate to the use of exogenous promoters. Applicants also argue that given the fact that Bronson provides for a random transgenesis method which is unspecific as to the site of integration and use of the promoter and Beach relating to random integration one of ordinary skill in the art would have not arrived to claimed invention. Applicants also argue that it is improper hindsight.

It appears that Applicant is arguing that the cited references do not expressly suggest the claimed invention of a method of gene knock down in a nonhuman

vertebrate. It is well established in case law that a reference must be considered not only for what it expressly teaches, but also for what it fairly suggests. In re Burkel, 201 USPQ 67 (CCPA 1979). Furthermore, in the determination of obviousness, the state of the art as well as the level of skill of those in the art is important factors to be considered. The teaching of the cited references must be viewed in light of these factors. It also appears that applicant is attempting to attack each reference individually. However, in a 103 rejection the references must be considered as a whole. In the instant case, McCaffrey/Beach teach a method of gene knock down in a nonhuman vertebrate by administering an expression vector comprising shRNA under the control of ubiquitous promoter that randomly integrates in the mouse genome. However, prior to filing of this application Bronson describes problems associated with DNA incorporated into the mouse germ line using this method includes random integration and unpredictable copy numbers. Bronson emphasize that random integration often also presents profound effect on expression of the transgene resulting in altered phenotype of the mouse (see page 9067, col.1, para. 1). In fact, Bronson provided advantages of targeting a single copy of a transgenic sequence to a chosen location in the genome such as HPRT over random integration of construct. He discloses many advantages of targeting at specific locus including the ability to control copy number, the ability to insert the transgene into regions of chromatin compatible with a desired developmental and tissue-specific expression. It is noted that Bronson et al emphasize that targeted transgenes provide a more efficient and informative means of securing and comparing the expression of various transgenic sequences than is available with current transgenic procedures.

Applicants err in stating that Bronson provides a method for a random transgenesis method which is unspecific as to the site of integration (see page 8 of the argument). In fact, contrary to applicants' assertion Bronson et al teach a method that uses homologous recombination in embryonic stem (ES) cells to generate mice having a single copy of a transgene integrated into a chosen location

in the genome. Specifically, Bronson et al disclose a method wherein a single copy murine bcl-2 cDNA driven by either a chicken beta-actin promoter or a human beta-actin promoter has been inserted immediately 5' to the HRPT locus by a directly selectable homologous recombination event (see the abstract and figure 2). Thus, given that method to stably integrate expression cassette at a chosen site in the genome was know in prior art. It would have been prima facie obvious for one of ordinary skill in the art to express the shRNA expression cassette that are flanked by homology regions for the endogenous loci such as hprt or rosa26 as described by Soriano et al. With respect to applicant's argument to the reference to Soriano et al, it appears that applicant's arguments focus on each reference individually. However, the test for combining references is not what the individual references themselves suggest, but rather what the combination of disclosures taken as a whole would have suggested to one of ordinary skill in the art. In re McLaughlin, 443 F.2d 1392, 170 USPQ 209 (CCPA 1971). For the purpose of combining references, those references need not explicitly suggest combining teachings, much less specific references. In re Nilssen, 7 USPQ2d 1500 (Fed. Cir. 1988). Soriano et al provided guidance with respect to ubiquitously expressed gene loci for use include Rosa 26, rosa5 and others (see col. 3, lines 49-54). Given that method that uses homologous recombination in embryonic stem (ES) cells to generate nonhuman animal having a single copy of a transgene integrated into a chosen location in the genome and advantage for targeted transgenes to attain more efficient expression was known in prior art, it would have been prima facie obvious for one of ordinary skill in the art to combine the teaching with reasonable expectation of success. Therefore, the claimed invention would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention.

New-Double Patenting

The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. A nonstatutory obviousness-type double patenting rejection is appropriate where the conflicting claims are not identical, but at least one examined application claim is not patentably distinct from the reference claim(s) because the examined application claim is either anticipated by, or would have been obvious over, the reference claim(s). See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); and *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) or 1.321(d) may be used to overcome an actual or provisional rejection based on a nonstatutory double patenting ground provided the conflicting application or patent either is shown to be commonly owned with this application, or claims an invention made as a result of activities undertaken within the scope of a joint research agreement.

Effective January 1, 1994, a registered attorney or agent of record may sign a terminal disclaimer. A terminal disclaimer signed by the assignee must fully comply with 37 CFR 3.73(b).

Claims 1, 5-6, 8-12, 14-24, 26-27 and 30 are provisionally rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claim14-16, 18-44 of copending Application No. 10/531,347. Although the conflicting claims are not identical, they are not patentably distinct from each other because claim 1 of the instant application is directed to a method for constitutive and/or inducible gene knock down in a non-human vertebrate selected from the group consisting of mouse and fish, which comprises stably integrating by homologous recombination an expression vector into a polymerase II dependent locus of the genome of the non-human vertebrate, said expression vector comprising a short hairpin RNA (shRNA) construct under control of a ubiquitous promoter and homologus sequences which integrate through homologous recombination at a polymerase II dependent locus of the genome of the non-human vertebrate, wherein the ubiquitous promoter is selected from the group consisting of polymerase I, II and III dependent promoters. Subsequent claims limit the polymerase II dependent

locus to include from a list elected from the group consisting of Rosa6, Hprt, collagen RNA polymerase. Claims are also directed to limit the vector containing functional sequences selected from the group consisting of splice acceptor sequences, polyadenylation sites and selectable marker sequences. Dependent claims 8-12, 14-19 limit the method of claim 1 to include various ubiquitous promoters. Claims 20-24 limits the shRNA to include a DNA segment A-B-C wherein A is a 15 to 35 bp DNA sequence with at least 95% complementarily to the gene to be knocked down; B is a spacer DNA sequence having 5 to 9 bp forming the loop of the expressed RNA hair pin molecule, and C is a 15 to 35 bp DNA sequence with at least 85% complementarily to the sequence A and further comprises a stop and/or polyadenylation sequence. Claim 27 is drawn to a a non-human vertebrate selected from the group consisting of mouse and fish having stably integrated by homologous recombination at a polymerase II dependent locus of the non-human vertebrate an expression vector comprising a short hairpin RNA (shRNA) construct under control of a ubiquitous promoter and homologous sequences which integrate at a polymerase II dependent locus of the genome of the non-human vertebrate, wherein the ubiquitous promoter is selected from the group consisting of polymerase I, II and III dependent promoters. Claim 30 is directed to an expression vector comprising a short hairpin RNA (shRNA) construct under control of a ubiquitous promoter and homologous sequences which integrate at a polymerase II dependent locus of the genome of a non- human vertebrate selected from the group consisting of mouse and fish, wherein the ubiquitous promoter is selected from the group consisting of polymerase I, II and III dependent promoters.. While claims 14-16 are directed to a method for constitutive and/or inducible gene knock down in a vertebrate, which method comprises stably integrating an expression vector comprising a short hairpin RNA construct under control of a ubiquitous promoter into the genome of the vertebrate, wherein the expression vector is suitable for stabile integration into the genome of a vertebrate, wherein the expression vector

contains homologous sequences suitable for integration at a defined genomic locus through homologous recombination in the genome of the vertebrate. Claims 18 and 19 limit the method to include homolgous sequences are suitable for integration at a polymerase II dependent locus in the genome of the vertebrate and expression vector further contains functional sequences selected from the group consisting of splice acceptor sequences, polyadenylation sites and selectable marker sequences. Claim 20 limits the polymerase II dependent locus is selected from the group consisting of a Rosa26, collagen, RNA polymerase, actin and HPRT locus. Claim 30 limits the method of to include vertebrate selected from the group of vertebrates consisting of mouse and fish. Subseqeunt claims limit the specific promoter and shRNA. Claims 41-43 are directed to a vertebrate having stably integrated, preferably at a polymerase II dependent locus of the vertebrate an expression vector comprising a short hairpin RNA construct under control of a ubiquitous promoters subsequently limiting vertebrates consisting of mouse and fish. Claim 44 is directed to an expression vector comprising a short hairpin RNA construct under control of a ubiquitous promoter. It is noted that certain of the instant claims differ only with respect to a narrower scope of vertebrate, promoter and construct, , which encompass those specifically claimed in application no 10/531,347.

This is a provisional obviousness-type double patenting rejection because the conflicting claims have not in fact been patented.

Conclusion

No Claims allowed.

The prior art made of record and not relied upon is considered pertinent to applicant's disclosure.Gossen et al (Proc Natl Acad Sci U S A. 1992 Jun 15;89(12):5547-51).

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Anoop Singh whose telephone number is (571) 272-3306. The examiner can normally be reached on 9:00AM-5:30PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Peter Paras can be reached on (571) 272- 4517. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

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